differences measured in the translocon experiments represent the differences in energy between the interfacial and the transmembrane conformations for a helix.

1113-Pos Board B23
Plasma Membrane Topology of Caveolins: Insights from Computational Modeling
Gene A. Morrill, Adele B. Kostellow.
Caveolins (CAV-1, CAV-2, CAV-3) are a family of integral membrane proteins, so-called because they are principal components of caveolae. They are known to facitate endocytosis and are involved in a number of biological functions, including signal transduction, cholesterol homeostasis, and transport. We find that the TOPCONS consensus sequence analysis algorithm as well as both the SVMtop and MEMSAT methods predict that each 22 kDa caveolin molecule contains two plasma membrane spanning helices (TM-1 and TM-2), separated by 1-2 amino acid residues at the membrane surface. Chem 3D Ultra visualization indicates that the transmembrane domains of all three caveolins are highly ordered α-helices. NetSurfP (Protein Surface Accessibility and Secondary Structure Predictions) analysis of the amino acid sequence further confirms the existence of two membrane spanning α-helices. These results conflict with the widely cited theory that caveolin forms a single 32 amino acid hydrophobic hairpin loop which is inserted into, but does not traverse, the plasma membrane. Based on our model, we find that TM-1,TM-2 and the interconnecting amino acid(s) in CAV-1, CAV-2 and CAV-3 of several vertebrate species are highly conserved. However, there are significant sequence differences between TM-1 and TM-2 of CAV-1 and CAV-2. The TMhit method, heptad repeat analysis and the coiled-coil prediction algorithm further indicate that helix-helix interactions occur both within and between these integral membrane proteins. The N-attachment (scaffold) domain also has a large intracellular helical content, contiguous with the TM-1 transmembrane helix. In contrast, the C-terminal attachment contains a disrupted helix, the first helical region being contiguous with transmembrane helix TM-2. The topology of the transmembrane helices described here is relevant to understanding caveolae formation and the mechanism of endocytosis as well as sequestration and release of membrane signaling molecules.

1114-Pos Board B24
Molecular Dynamic Simulations of Apolipoprotein A-I peptide Mimetics
Amanda M. Plain, James C. Patterson.
Coronary heart disease is the leading cause of death in the United States, claiming more lives than the next seven leading causes of death combined. High levels of high density lipoprotein (HDL) have been correlated with lower rates of coronary heart disease. Apolipoprotein A-I (apoA-I), is the principle protein in HDL, is a 243-residue class A amphipathic alpha helix capable of binding a variable number of lipid molecules. ApoA-I mimetic peptides synthesized by Anantharamaiah et al. are 18-residue class A amphipathic helices. Although they have no sequence homology to apoA-I, the peptides bind to lipids in a manner similar to that of apoA-I (i.e. antiparallel double-belt on the edge of the lipid disc).
Molecular dynamics simulations of the lipid-bound peptide mimetic 2F, $3F^{2}$ and $3F^{14}$ were performed for a minimum of 30 ns in explicit water using CHARMM22/27 parameters. The peptides were arranged in a double belt conformation around the lipid. In the model, 16 straight alpha-helical chains of each peptide were placed around two leaflets of 108 dimyristoylphosphatidylcholines (DMPC). Each simulation became saddle shaped around the lipids. The complexes remained intact throughout the simulation. Each peptide maintained a similar number of hydrogen bonds with the lipids. The 4F peptides cause the bilayer to pinch more than the other peptides that were explored. The interaction energies of the peptides are similar, as are the interchain interaction energies.

1115-Pos Board B25
Multiscale Simulations of Lipid Interactions with Integral Membrane Proteins: Aquaporins
Phillip J. Stansfeld, Elizabeth E. Jefferys, **Mark S.P. Sansom**.
Membrane protein structural biology is one of the key biochemical challenges of the coming decade. With continuous improvements to the methods used by structural biologists there is a predicted exponential growth in the number of membrane proteins structures. Nevertheless, these biological assemblies are usually resolved in the absence of the native lipid environment. Coarse-Grained molecular dynamics (CGMD) simulations provide a means for assessing the assembly and interactions of molecular complexes at a reduced level of representation. This method has been shown to accurately predict the insertion position of proteins within a cell membrane. Previous studies enabled us to correlate predicted contacts to lipids with experimental data on lipid-exposed residues for a number of membrane proteins (e.g. LacY, rhodopsin, KcsA, MscL, FepA, BtuB). The recent determination of high resolution structures for aquaporins in a phospholipid bilayer environment (Aqp0: 2B6O and 3M9I; Aqp4: 2ZZ9) has provided an excellent test case. The CGMD approach has been used to simulate lipid/protein interactions for >30 members of the aquaporin family, revealing that patterns of protein/lipid headgroup interactions are conserved and are in good agreement with the lipids resolved in the electron crystallography structures. We have extended this further by using a multi-scale approach, investigating the interactions at atomic resolution. At this scale we may also consider hydrogen bonding interactions, which also show good agreement with the experimental structures. This methodology is being incorporated into a semi-automated high-throughput pipeline to enrich our database of CGMD membrane protein simulations (CGDB; http://sbcb.bioch.ox.ac.uk/cgdb).

1116-Pos Board B26
Comparative Neural Network and Alignment Study of Aquaporins
Julie Bomholt, Jesper S. Hansen, Per Greisen Jr., Dan A. Klærke, Claus Hélix-Nielsen.
Common for the membrane channel protein family of aquaporins is that they facilitate a rapid transport of water molecules. Aquaporins are found in a wide range of organisms and several protein structures have been solved. Although they share structural similarities there are also distinct features for each class (e.g. plant versus mammalian aquaporins). This raises the question: How well does sequence and structural features of the respective proteins correlate to functional sites? We here present a comparative study of aquaporins based on neural network analysis as well as structure, sequence and residue alignments. Using HotPatch, an automated algorithm developed by Pettit et al. (Pettit, F. K. et al. J. Mol. Biol. 2007 369, 863-879) we calculated probable aquaporin functional interaction sites such as sites involved in (specific) lipid interactions. Alignment of the calculated sequences allowed us to analyze similarities and differences in the proteins in a quantitative manner as well as elucidating the potential functional role of specific residues. This will further give us new information on functionally important features that could assist in the design of proteins with increased stability for biomimetic applications.

1117-Pos Board B27
Small-World and Random Networks in Contact Maps of Protein Channels
Malgorzata Kotulska, Mark B. Gerstein.
Although it is estimated that protein channels constitute large portion of all proteins, the number of their experimental structures is extremely small compared to other proteins. Lipid environment and great size of the channels impede the development. Improvement could contribute to drug design and resolving mechanisms of channelopathies. Modeling 3D structure of transmembrane channels can be enhanced by constraints defined by their contact sites distribution, supporting proper selection of protein decoys in the modeling process. We represent a protein as a network of contact sites and characterize topology of the graph. As a contact site we consider residues within a defined distance between protein backbone C alpha (or beta) atoms that are separated by a certain number of amino acids. Characteristics of the graph model is tested, with regard to protein secondary structure and location of the residues in the membrane or outside. We investigate how the topological characteristics of protein channels compare to other proteins and how definition of the contact site, in terms of the distance and separation threshold values, affects the network topology. The results indicate that the graph model of ionic channel has distinct features. The network topology of proteins proved sensitive to the contact site definition, showing that the graph obeys typical real world network characteristics (“small-world” graph) only if the distance and the separation in the contact site definition are within certain limits, which are not always in accordance with typically assumed. Otherwise a random (Erdos-Rényi) network is observed. Transmembrane helices are much less sensitive to the contact site definition and fall into the “small world” more easily than other structures.